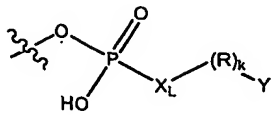
WHAT IS CLAIMED IS:

1. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide comprising a group having the formula



wherein:

X_L is selected from the group consisting of NH, NR_1 and O, wherein R_1 is selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl or heterocycloalkyl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, $-NR_2CO-$. $-COOR_2-$, -CONH-, and $-CONHR_2-$ groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

2. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl, heteroaryl, -CO-, -NHCO-, -O-, -S-, $-NR_2CO-$. $-COOR_2-$, -CONH-, and $-CONHR_2-$ group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group.

3. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein each R group is a bifunctional substituent independently

selected from the group consisting of unsubstituted heteroalkyl, alkyl, cycloalkyl, heterocycloalkyl, -CONH-, -NHCO- and -CO- groups.

4. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein X_L is NH.

5. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 the X_L is O.

6. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein X_L is NR_1, wherein R_1 is selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, or heterocycloalkyl group.

7. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein k is a whole number from 1-10.

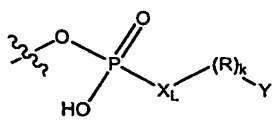
8. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein Y is selected from the group consisting of biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein and beta-lactamase.

9. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 8 wherein Y is Biotin/Avidin.

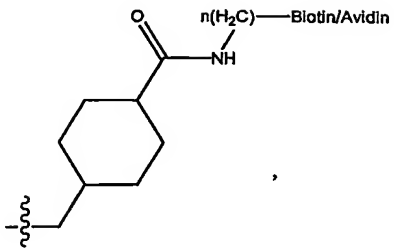
10. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein X is NH, Y is Biotin/Avidin, and each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted heteroalkyl, alkyl, cycloalkyl, heterocycloalkyl, -CONH-, -NHCO- and -CO- groups.

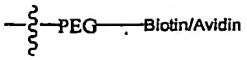
11. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein X is NH, Y is Biotin/Avidin, each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted heteroalkyl, alkyl, cycloalkyl, heterocycloalkyl, -CONH-, -NHCO- and -CO- groups, and k is a whole number from 1-14.

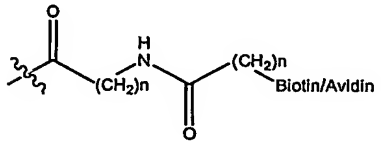
12. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 wherein the $-(R)_K-Y$ moiety of the group of the formula



is selected from the group consisting of







and

Where n is a whole number from 1-25.

13. A polypeptide according to claim 1.

14. A polypeptide according to claim 9.

15. A polypeptide according to claim 11

16. An oligonucleotide according to claim 1.

17. A lipid according to claim 1.

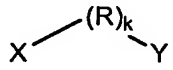
18. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 1 further comprising a label.

19. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 11 further comprising a label.

20. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 18 wherein the label is a fluorophore.

21. A polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide according to claim 20 wherein the fluorophore is StarBright®.

22. A method of covalently attaching one or more hetero-bifunctional linkers having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl,

heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group;

to one or more hydroxyl moiety of a polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide comprising the steps of:

(a) converting the hydroxyl moiety to phosphoric acid or amino derivative of phosphoric acid; and

(b) converting the phosphoric acid or amino derivative of phosphoirc acid to a phosphoro analog by reacting the phosphoric acid or amino derivative of phosphoric acid with the hetero-bifunctional linker.

23. A method according to claim 22 wherein the phosphoryl analog is a phosphoramidate, phosphoroamino or phosphorothiol analog.

24. A method according to claim 22 wherein the hydroxyl moiety is on a polypeptide containing one or more free amine or carboxylic acid group wherein the free amine or carboxylic acid group is protected before step (a).

25. A method according to claim 22 wherein the phosphoryl analog contains an amino, carboxyl or sulfhydryl group covalently attached to a detectable label.

26. A method according to claim 25 wherein the detectable label is a fluorphore.

27. A method according to claim 25 wherein the detectable label is covalently attached by reacting the label with either a homo-bifunctional linker or a hetero-bifunctional linker.

28. A method according to claim 22 wherein the hydroxyl moiety is a 5' hydroxyl group on a oligonucleotide, nucleoside or nucleotide.

29. A method according to claim 28 wherein ATP or [γ-NH_2]-ATP is used in step (a) to convert the hydroxyl moiety to the phosphoric acid or amino derivative of phosphoric acid.

30. A method according to claim 22 wherein the hydroxyl moiety is present on a tyrosine, serine, threonine or histadine group.

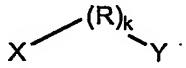
31. A method according to claim 30 wherein ATP or [γ-NH_2]-ATP is used in step (a) to convert the hydroxyl moiety to the phosphoric acid or amino derivative of phosphoric acid.

32. A method according to claim 22 wherein the polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide comprises a detectable label.

33. A method according to claim 32 wherein the detectable label is a fluorphore.

34. A method according to claim 22 wherein the phosphoric acid derivative is converted into a more reactive intermediate before it is reacted with the hetero-bifunctional linker to form the phosphoro analog.

35. A method of covalently attaching one or more hetero-bifunctional linkers having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold,

polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group;

to a hydroxyl moiety of a polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide comprising the steps of:

(a) converting the hydroxyl moiety to a phosphoric acid derivative; and

(b) converting the phosphoric acid derivative to a phosphoro analog by reacting the phosphoric acid derivative with a preformed nucleation reagent comprising a hetero-bifunctional linker.

36. A method according to claim 35 wherein the phosphoryl analog is a phosphoramidate, phosphoroamino or phosphorothioate analog.

37. A method according to claim 35 wherein the hydroxyl moiety is on a polypeptide containing one or more free amine or carboxylic acid groups wherein the free amine or carboxylic acid groups are protected before step (a).

38. A method according to claim 35 wherein the phosphoryl analog contains an amino, carboxyl or sulfhydryl group covalently attached to a detectable label.

39. A method according to claim 35 wherein the detectable label is a fluorophore.

40. A method according to claim 35 wherein the detectable label is covalently attached by reacting the label with either a homo-bifunctional linker or a hetero-bifunctional linker.

41. A method according to claim 35 wherein the hydroxyl moiety is a 5' hydroxyl group on a oligonucleotide, nucleoside or nucleotide.

42. A method according to claim 41 wherein ATP, [γ-S]-ATP or [γ-NH_2]-ATP is used in step (a) to convert the hydroxyl moiety to the phosphoric acid derivative.

43. A method according to claim 35 wherein the hydroxyl moiety is present on a tyrosine, serine, threonine or histamine group.

44. A method according to claim 43 wherein ATP, [γ-S]-ATP or [γ-NH_2]-ATP is used in step (a) to convert the hydroxyl moiety to the phosphoric acid derivative.

45. A method according to claim 35 wherein the preformed nucleation reagent comprises one or more high molecular weight reagent conjugated to one or more hetero-bifunctional linker.

46. A method according to claim 45 wherein the high molecular weight reagent is selected from oligomeric polymers, synthetic polymers, and mixed co-polymer derivatives.

47. A method according to claim 45 wherein the high molecular weight reagent is selected from polyamino acids, polypeptides, polylysine, poly acrylic acid, polyamines, and polysulfones.

48. A method according to claim 45 wherein the heterobifunctional linker is succinimidyl-4-(N-maleimidomethyl)cyclohexane-1-carboxy-6-amidocaproate or succinimidyl-3-(bromoacetamido)propionate.

49. A method according to claim 45 wherein Y of the hetero-bifunctional linker is biotin and the high molecular weight reagent is avidin.

50. A method according to claim 35 wherein the preformed nucleation reagent comprises a fluorophore.

51. A method according to claim 35 wherein the preformed nucleation reagent comprises a reactive amine group and a biotin/avidin complex.

52. A method according to claim 35 wherein the polypeptide, lipid, amino acid, nucleoside, nucleotide or oligonucleotide comprises a label.

53. A method according to claim 53 wherein the label is a fluorophore.

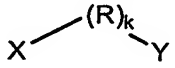
54. A method according to claim 35 wherein the phosphoric acid derivative is converted into a more reactive intermediate before it is reacted with the nucleation reagent to form the phosphoro analog.

55. An assay for detecting phosphorylation or de-phosphorylation comprising:

(a) a kinase substrate having one or more hydroxyl moiety;

(b) a kinase enzyme and ATP or [γ-NH_2]-ATP; and

(c) a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

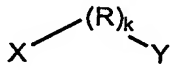
Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

56. An assay for detecting phosphorylation or de-phosphorylation comprising:

(a) a phosphatase substrate having one or more hydroxyl moiety;

(b) a phosphatase enzyme; and

(c) a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

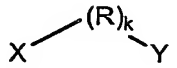
57. A homogeneous assay for detecting phosphorylation or de-phosphorylation comprising:

(a) a polypeptide substrate having one or more hydroxyl moiety;

(b) a protein kinase enzyme and ATP or [γ-NH_2]-ATP;

(c) a monomeric high molecular weight reagent; and

(d) a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

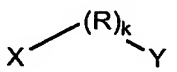
58. A homogeneous assay for detecting phosphorylation or de-phosphorylation comprising:

(a) a polypeptide substrate having one or more hydroxyl moiety;

(b) a protein phosphatase enzyme;

(c) a monomeric high molecular weight reagent; and

(d) a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, $-NR_2CO-$. $-COOR_2-$, -CONH-, and $-CONHR_2-$ groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

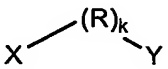
59. A homogeneous assay for detecting phosphorylation or de-phosphorylation comprising:

(a) a polypeptide substrate having one or more hydroxyl moiety;

(b) a protein kinase enzyme, and ATP an ATP analog or any other phosphate donating group; and

(c) a preformed nucleation reagent.

60. A homogeneous assay according to claim 59 wherein the preformed nucleation reagent is formed by attaching one or more high molecular weight reagent to one or more hetero-bifunctional linkers having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

61. A homogeneous assay according to claim 60 wherein the hetero-bifunctional linker has a reactive amino terminus.

62. A homogeneous assay according to claim 60 wherein the high molecular weight reagent is selected from oligomeric polymers, synthetic polymers, mixed co-polymer derivatives, polyamino acids, polypeptides, polylysine, poly acrylic acid, polyamines and polysulfones.

63. A homogeneous assay according to claim 60 wherein the high molecular weight reagent is avidin and the hetero-bifunctional linker comprises biotin.

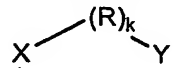
64. A homogeneous assay for detecting phosphorylation or de-phosphorylation comprising:

(a) a polypeptide substrate having one or more hydroxyl moiety;

(b) a protein phosphatase enzyme; and

(c) a preformed nucleation reagent.

65. A homogeneous assay according to claim 64 wherein the preformed nucleation reagent is formed by attaching one or more high molecular weight reagents with one or more hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected

from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

66. A homogeneous assay according to claim 65 wherein the X group of the hetero-bifunctional linker is an amine.

67. A homogeneous assay according to claim 66 wherein the high molecular weight reagent is avidin and ther hetero-bifunctional reagent further comprises biotin.

68. A homogeneous assay according to claim 65 wherein the high molecular weight reagent is selected from oligomeric polymers, synthetic polymers, mixed co-polymer derivatives, polyamino acids, polypeptides, polylysine, poly acrylic acid, polyamines, and polysulfones.

69. A assay for detecting phosphorylation or de-phosphorylation comprising the steps of:

(a) attaching a fluorophore to a polypeptide;

(b) blocking at least one reactive free amine or acid group on the polypeptide to form a protected polypeptide;

(c) phosphorylating the protected polypeptide with ATP, an ATP analog or any other phosphate donating substrate; and

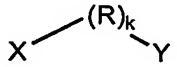
(d) measuring fluorescence polarization.

70. A method for blocking one or more reactive amine or acid groups on a polypeptide target comprising:

(a) treating the polypeptide target with a fluorescein-NHS or StarBright® Dye Isothiocyanate; and then

(b) protecting one or more free amines or acids present on the polypeptide target.

71. A method of preparing a preformed nucleation center comprising the step of reacting one or more high molecular weight reagent with one or more hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

72. A method according to claim 71 wherein the high molecular weight reagent is selected from oligomeric polymers, synthetic polymers, mixed co-polymer derivatives, polyamino acids, polypeptides, polylysine, poly acrylic acid, polyamines and polysulfones.

73. A method according to claim 71 wherein the hetero-bifunctional linker has a reactive amino group.

74. A method according to claim 71 wherein the high molecular weight reagent is avidin and the hetero-bifunctional linker comprises biotin.

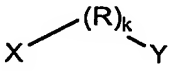
75. A method according to claim 74 wherein the hetero-bifunctional linker further comprises a reactive amino group.

76. An assay for the detection of phosphorylation and de-phosphorylation comprising:

(a) combining a protein kinase, one or more test compounds, a labeled polypeptide substrate, and ATP or [γ-NH_2]-ATP an ATP to make a first mixture;

(b) allowing the first mixture to react to form a phosphopeptide product;

(c) adding a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group;

to create a phosphorylated polypeptide product; and

(d) measuring the fluorescence polarization of the phosphorylated polypeptide product.

77. An assay according to claim 76 wherein the Y group of the hetero-bifunctional linker is biotin.

78. An assay according to claim 76 wherein avidin is added to the phosphorylated polypeptide product before step (d).

79. An assay according to claim 76 wherein the hetero-bifunctional linker is present in a preformed nucleation mixture comprising the hetero-bifunctional linker and a high molecular weight reagent.

80. An assay according to claim 68 wherein the hetero-bifunctional linker covalently binds to an amine group of the phosphopeptide product to form the phosphorylated polypeptide product.

81. An assay according to claim 76 wherein the labeled polypeptide substrate is labeled with a fluorphore.

82. An assay according to claim 76 wherein the phosphopeptide product is formed in less than three hours.

83. An assay according to claim 82 wherein the phosphopeptide product is formed in less than one hours.

84. An assay according to claim 83 wherein the phosphopeptide product is formed in less than 30 minutes.

85. An kit comprising the materials for the assay of claim 76.

86. A kit according to claim 85 in a configuration selected from microtiter plates, fluorescence microwell plate readers, microfluic devices, bead-based separatory devices, fluorescence microscopes and electrophoresis gels.

87. An assay for the detection of phosphorylation and de-phosphorylation comprising:

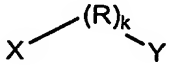
(a) combining a protein kinase, one or more test compounds, a labeled polypeptide substrate, and ATP or an ATP analog to make a first mixture;

(b) allowing the first mixture to react to form a phosphopeptide product;

(c) adding a preformed nucleation reagent to create a phosphorylated polypeptide product; and

(d) measuring the fluorescence polarization of the phosphorylated polypeptide product.

88. An assay according to claim 87 wherein the preformed nucleation reagent comprises a high molecular weight reagent associated with a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

89. An assay according to claim 88 wherein Y of the hetero-bifunctional linker is biotin and the high molecular weight reagent is avidin.

90. An assay according to claim 88 wherein the hetero-bifunctional linker covalently binds to an amine or thio group of the phosphopeptide product to form the phosphorylated polypeptide product.

91. An assay according to claim 87 wherein the ATP analog is [γ-S]-ATP or [γ-NH_2]-ATP.

92. An assay according to claim 87 wherein the labeled polypeptide substrate is labeled with A fluorphore.

93. An assay according to claim 87 wherein the phosphopeptide product is formed in less than three hours.

94. An assay according to claim 93herein the phosphopeptide product is formed in less than one hour.

95. An assay according to claim 94 wherein the phosphopeptide product is formed in less than 30 minutes.

96. An kit comprising the materials for the assay of claim 87.

97. A kit according to claim 96 in a configuration selected from microtiter plates, fluorescence microwell plate readers, microfluic devices, bead-based separatory devices, fluorescence microscopes and electrophoresis gels.

98. A kit for the detection of phosphorylation or de-phosphorylation comprising:

(a) a target substrate;

(b) a reaction buffer;

(c) an enzyme;

(d) ATP or an ATP analog; and

(e) a nucleation reagent.

99. A kit according to claim 98 wherein the target substrate is a polypeptide target.

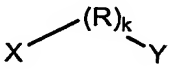
100. A kit according to claim 99 wherein the polypeptide target is from 5-500 amino acids.

101. A kit according to claim 100 wherein the ATP analog is [γ-S]-ATP or [γ-NH_2]-ATP.

102. A kit according to claim 98 wherein the target substrate is labeled.

103. A kit according to claim 102 wherein the target substrate is labeled with a fluorphore.

104. A kit according to claim 98 wherein the nucleation reagent comprises a high molecular weight reagent and a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$-group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

105. A kit according to claim 104 wherein the high molecular weight reagent is avidin.

106. A kit according to claim 104 wherein the high molecular weight reagent and the hetero-bifunctional linker are stored separately.

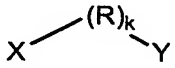
107. A kit according to claim 98 wherein the target substrate, reaction buffer, enzyme, ATP or ATP analog, and nucleation reagent are kept separate from each other until used.

108. A kit according to claim 98 in a homogeneus assay configuration selected from microtiter plates, fluorescence microwell plate readers, microfluid devices, bead-separatory devices, and electrophoresis gels.

109. A preformed nucleation reagent comprising:

(a) a high molecular weight reagent; associated with

(b) a hetero-bifunctional linker having the formula



wherein:

X is selected from the group consisting of NH_2, succinimidyl, maleimidyl, iodoacetamido, bromoacetamido, thiol, SO_2, SO, and NHR_2 group; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

each R group is a bifunctional substituent independently selected from the group consisting of unsubstituted or substituted alkyl, heteroalkyl, aryl, heteroaryl, cycloalkyl, heterocycloalkyl, -CO-, -NHCO-, -O-, -S-, -NR_2CO-. -$COOR_2$-, -CONH-, and -$CONHR_2$- groups; wherein R_2 is selected from hydrogen and unsubstituted alkyl, heteroalkyl, cycloalkyl, heterocycloalkyl, aryl and heteroaryl group;

k is a whole number from 1 to 100; and

Y is selected from from the group consisting of biotin, biotin/avidin, biotin/streptavidin, alkaline phosphatase, casein, beta-lactamase, Bovine Serum Albumin, IgG, Avidin-alkaline phosphatase, streptavin-alkaline phosphatase, and biotin or streptavidin complexed with one or more group selected from glycoproteins, enzymes, antibodies, nucleotides, polypeptides, derivatized particles made of polystyrene, nylon, gold, polyacrylamide, and solid surfaces selected from microtiter pates, silicon plates, and polymers comprising at least one active functional group.

110. A preformed nucleation reagent according to claim 109 wherein the high molecular weight reagent is selected from oligomeric polymers, synthetic polymers, mixed co-polymer derivatives, polyamino acids, polypeptides, polylysine, poly acrylic acid, polyamines, and polysulfones.

111. A preformed nucleation reagent according to claim 109 wherein the X group of the hetero-bifunctional linker is an amine.

112. A preformed nucleation reagent according to claim 109 wherein the high molecular weight reagent is avidin and Y on the hetero-bifunctional linker is biotin.

113. A method according to claim 109 wherein the hetero-bifunctional linker comprises a reactive amino group.

114. A compound of the formula